

## Review

# The temporary effect of weed-cover maintenance on transpiration and carbon assimilation of olive trees

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## ABSTRACT

The maintenance of spontaneous weed cover is a conservation practice used in olive groves. Herbaceous plants in alleys between the trees can increase the capacity of this agroecosystem to remove carbon. However, the influence of this practice on carbon assimilation at the leaf scale has not yet been studied in olive trees. Also, the presence of other species competing with olive trees for soil water has the potential to modify the water use efficiency, a key parameter in a climate change context. In this study, leaf-scale net carbon assimilation ( $A_{\text{leaf}}$ ), transpiration ( $E_{\text{leaf}}$ ) and water use efficiency as the ratio  $A_{\text{leaf}}/E_{\text{leaf}}$  ( $WUE_{\text{leaf}}$ ) were quantified in olive grove divided by two different treatments: (1) a weed-free (WF) ecosystem in which weed growth was inhibited by applying herbicide; and (2) a weed-covered (WC) ecosystem in which spontaneous herbaceous plants were kept and then mechanically mowed and left on the ground. A portable leaf photosynthesis system was used to measure olive leaf fluxes for both treatments, and likewise for the ecosystem scale via two eddy covariance towers assessing gross primary production ( $GPP_{\text{eco}}$ ), evapotranspiration ( $ET_{\text{eco}}$ ), and water use efficiency ( $WUE_{\text{eco}}$ ). We found that the average  $A_{\text{leaf}}$  was 24% higher in the WF treatment while  $GPP_{\text{eco}}$  decreased 32% compared to WC treatment. However,  $A_{\text{leaf}}$  was significantly different between treatments only during weed growth: January-May ( $A_{\text{leaf-WF}} = 7.6 \pm 3.7 \mu\text{mol CO}_2\text{m}^{-2}\text{s}^{-1}$ ;  $A_{\text{leaf-WC}} = 5.1 \pm 3.1 \mu\text{mol CO}_2\text{m}^{-2}\text{s}^{-1}$ ) while  $A_{\text{leaf}}$  was similar between the two treatments after mowing. Mowed weeds decreased  $T_{\text{soil}}$  and VPD, and these changes were accompanied by a decrease in  $E_{\text{leaf}}$  in olive trees. Therefore, this led to  $WUE_{\text{leaf-WF}} > WUE_{\text{leaf-WC}}$  when the weeds were growing and the opposite after mowing. Thus, although the presence of spontaneous weeds increased the annual ecosystem C uptake in the olive orchard, both  $A_{\text{leaf}}$  and seasonal fluctuations in  $WUE_{\text{leaf}}$  were reduced with weed maintenance.

## 1. Introduction

Accelerated soil erosion entails an increase of atmospheric  $\text{CO}_2$  emissions and is a growing problem that requires adaptive management towards the re-carbonization of soils (Lal, 2019). Crop management can result in storing atmospheric carbon in agricultural soils (Aguilera, 2013; Almagro et al., 2016), which helps mitigate climate change. These practices can be especially beneficial in Mediterranean climates with low soil carbon content, typically below  $< 15 \text{ g C kg}^{-1}$  in croplands (de Brogniez et al., 2015; Rodríguez Martín et al., 2016). Climatic conditions of the Mediterranean region are characterized by irregular rainfall, high demand for evaporation, and frequent water shortages. When soil is

devoid of vegetation due to herbicides or regular tillage, these climatic conditions facilitate soil erosion (García-Ruiz et al., 2013).

Maintaining spontaneous weed cover in the alleys of tree crops is a frequent soil conservation method with several advantages. The cover considerably reduces erosion, increases nutrient retention after rain events, and transfers atmospheric  $\text{CO}_2$  to soil through their roots (González-Sánchez, 2012; Aguilera et al., 2015; Poeplau, 2015; Martínez-Mena et al., 2020). Additionally, this practice may increase water infiltration and therefore the soil water content (Basche and DeLonge, 2019), despite weed competition for water (Abouzienda et al., 2015). Porosity also increases and therefore field capacity, improving soil quality and enhancing the sustainability of crop production systems

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especially in dry environments (Basche and DeLonge, 2017; Haruna et al., 2018). Weeds provide many other benefits such as improved physicochemical and microbial soil quality (Ramos et al., 2011; Blanco-Canqui et al., 2015), and increased biodiversity (Sokos et al., 2013; Schmidt et al., 2019; Kim et al., 2020), seed dispersal and pollinators (Baraibar et al., 2011; Nicholls and Altieri, 2013; Lee et al., 2019).

Weed cover is a commonly used conservation practice in olive trees (*Olea europaea* L.), which are among the most representative tree crops in the Mediterranean basin. Consequently, their management has enormous economic, social and environmental implications for this region that represents >95% of world production (FAOSTAT, 2019). After establishment of a weed cover, erosion rates are notably reduced in olive groves and soil carbon content increases considerably in addition to other nutrients (Gómez et al., 2004; Castro et al., 2008; Martínez-Mena et al., 2008; Gómez et al., 2009; Marquez-García et al., 2013; Nieto et al., 2013; Palese et al., 2013; Repullo-Ruibérriz et al., 2018; Sastre et al., 2018; Martínez-Mena et al., 2020). Olive groves usually have a positive global warming potential that can be neutralized in organic olive groves (Aguilera et al., 2015). The meta-analyses of Aguilera et al. (2013), Vicente-Vicente et al. (2016) and Morugán-Coronado et al. (2020) show increased carbon sequestration when maintaining weed cover in this agroecosystem. However, although such practices are expanding, they generate controversy in semi-arid environments (Pastor et al., 2001; Morugán-Coronado et al., 2020) because farmers do not observe consistent improvements in yields (Alcantara et al., 2011). Furthermore, the presence of weeds can reduce crop yield due to competition for water (Slaughter et al. 2008) and while a mown weed mulch could reduce soil evaporation (Connor, 2005), living herbaceous plants may compete with the main crop for water, modify the environmental conditions and therefore induce changes in leaf-level exchanges of water vapor and CO<sub>2</sub>. During long periods of drought, olive trees reduce their photosynthesis and, therefore, their productivity (Moriana et al., 2003; Ben Ahmed et al., 2007). To solve this problem irrigation is usually applied in semi-arid environments.

A better understanding of atmosphere-ecosystem carbon exchange mechanisms can facilitate interventions capable of improving C capture (Marland et al., 2004). Although net ecosystem exchange (NEE) of CO<sub>2</sub> in olive groves has been quantified by destructive measures (Sofo et al., 2005; Villalobos et al., 2006), the eddy covariance (EC) technique has become an important tool in recent years since it allows assessing NEE at large spatial and temporal scales without altering the studied ecosystem (Baldocchi, 2003; Baldocchi et al., 2020). Using this technique Nardino et al. (2013) demonstrated a high capacity of young olive trees as carbon sinks. Brilli et al. (2016) concluded that olive trees with conventional management are important carbon sinks. Testi et al. (2008) and López-Bernal et al. (2015) also used this technique to measure CO<sub>2</sub> exchanges in olive trees, but the data record did not reach one year and, as in the above cases, the influence of a weed cover was not considered. To date, only one study has quantified the effect of weed cover on the net carbon balance at ecosystem level, showing that a covered olive ecosystem sequesters twice as much carbon as an ecosystem without weed cover (Chamizo et al., 2017). However, the cited study did not quantify differences in the behaviour of the carbon and water fluxes of the olive trees under the two treatments. Due to the important contribution of weed cover detected in the CO<sub>2</sub> fluxes at the ecosystem level, a more in-depth study of the effect of weed cover on the different compartments of these agroecosystems is essential.

To determine if carbon assimilation of olive trees is affected by the establishment of weed cover, it is necessary to obtain gas fluxes at a smaller spatial scale. In this sense, CO<sub>2</sub> fluxes have been sampled at plant level with chamber systems (Villalobos et al., 2012; Pérez-Priego et al., 2014) characterizing the assimilation of olive trees, or at the ground level (Bertolla et al., 2014; Sierra et al., 2016; Turrini et al., 2017) showing that olive groves with weed cover had higher rates of soil respiration. At the leaf scale, net CO<sub>2</sub> assimilation ( $A_{\text{leaf}}$ ) has been well characterized in olive trees (Higgins et al., 1992; Proietti and Famiani,

2002; Larbi et al., 2015; Bedbabis et al., 2017), but there are no studies investigating the effect of the weed cover on the  $A_{\text{leaf}}$  of the olive trees.

In the context of the water balance in olive groves, it is known that transpiration is the main process in olive groves when the soil is dry and that evaporation is important only after rainfall events (Testi et al., 2004). Since drier conditions are predicted in the Mediterranean (Martiotti et al., 2015), evapotranspiration ( $ET_{\text{eco}}$ ) is expected to increase in irrigated olive groves by an average of 8% by mid-century (Tanasijevic et al., 2014) modifying the instantaneous water use efficiency (WUE: ratio of carbon assimilation to transpiration), a key metric to evaluate the functioning of the agroecosystem in response to different environmental conditions (Boese et al., 2019). In this regard, the study of  $ET_{\text{eco}}$  and WUE in olive groves under different treatments is crucial. To date,  $ET_{\text{eco}}$  in olive groves has been directly quantified by applying water balances (Palomo et al., 2002), models based on crop coefficient or on stomatal conductance (Orgaz et al., 2006; Orgaz et al., 2007; Paço et al., 2014), semi-empirical equations (Allen et al., 1998; Droogers, 2000), sap flow measurements (Ayyoub et al., 2017), remote sensing data (Aguirre-García et al., 2021) or the EC technique (Testi et al., 2004; Chebbi et al., 2018). However, the most promising techniques, such as remote sensing or EC, estimate  $ET_{\text{eco}}$  at the ecosystem level and must model or quantify separately the contribution of each process and organ, such as leaf transpiration ( $E_{\text{leaf}}$ ). So far, of the many studies carried out in olive groves, none have focused on studying the effect of weed cover on  $E_{\text{leaf}}$  or leaf-level WUE ( $WUE_{\text{leaf}}$ ).

Although weed cover can lead to changes in carbon and water fluxes at the ecosystem level, its effect at the crop leaf level is unknown. Since in other Mediterranean crops, like vineyards, the interactions between weeds and the crop depends on weed density (Novara et al., 2021), we hypothesize that, under full weed development and with the same irrigation, the presence of weeds may modify leaf-scale fluxes in olive trees by changing soil and air temperature and humidity, reflected radiation and nutrients available to the olive tree. Therefore, the main objectives of this study were to i) quantify the seasonal variation in CO<sub>2</sub> and water vapor fluxes and WUE at leaf level in two irrigated olive groves, one with weed cover (WC) in the alleys and the other weed free (WF), and determine if weed cover alters these fluxes, and ii) analyse whether there is different behaviour between treatments in gas exchanges at leaf and ecosystem scales. To address these objectives, we analysed monthly measurements of leaf-level and ecosystem fluxes in an olive grove in the south of Spain through a year.

## 2. Material and methods

### 2.1. Site description

This study was carried out in an irrigated olive grove (*Olea europaea* L. "Arbequina") from "Cortijo Guadiana" (37°5445N; 3°1340W; 370 m.a.s.l.), in Úbeda (Jaén, Spain) that belongs to the olive oil company Castillo de Canena, SL (Fig. 1). The climate is Mediterranean (Csa; Köppen classification) with dry and warm summers. Mean annual temperature is 16 °C. Annual precipitation is 470 ± 160 mm and potential evapotranspiration 1205 ± 95 mm (n = 18; IFAPA, 2020). From April to October trees were drip-irrigated three times a week at night with a total contribution of 135 mm. Trees are on clay loam soil and were ferti-irrigated with the addition of 25-40 g of NPK fertilizer per tree each night. Tree height is ca. 4 m and age 85 years. The plantation frame is 12×12 m, which means a distribution of 69-70 trees / ha and 27% tree ground cover.

In 2014, two homogeneous and flat parcels of the olive grove were selected to apply different treatments: 1) weed free (WF), with glyphosate-based herbicide applied in fall (September 2017) and winter (February 2018) to prevent plant growth and 2) weed covered (WC), with weeds growing naturally from autumn and eventually occupying 100% of the soil. In spring (June 10<sup>th</sup> and 11<sup>th</sup>, 2018) weeds were mechanically mowed and kept on the soil until natural degradation. Leaf

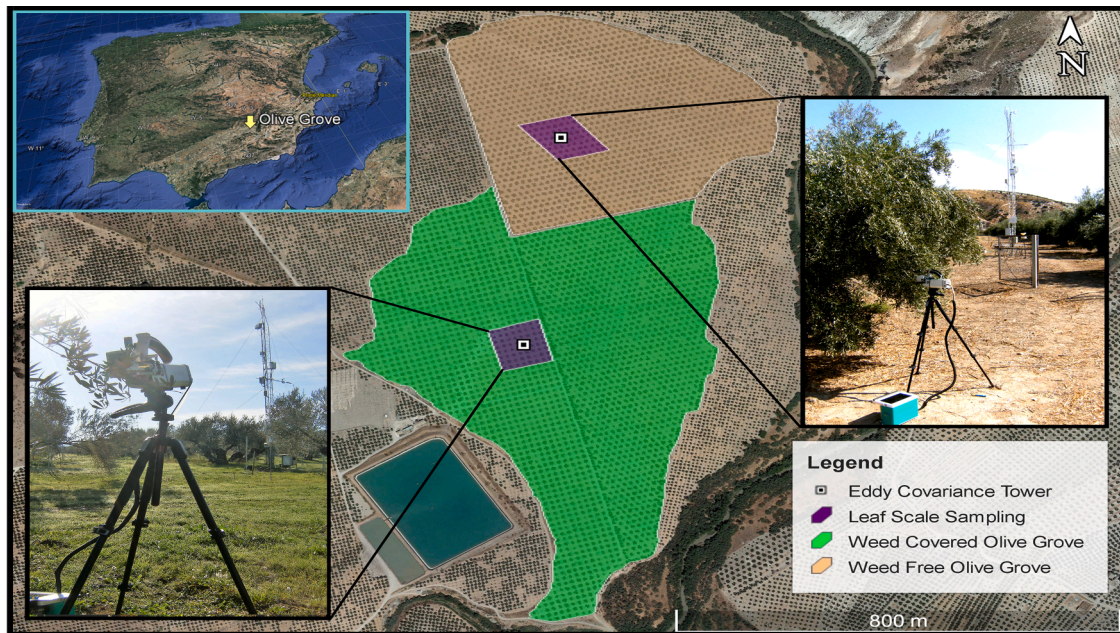


Fig. 1. Location of olive grove, leaf scale sampling area and surface of each treatment of the experimental site: keeping of spontaneous weeds (weed covered; WC) and removal of spontaneous weeds after herbicide application (weed free; WF). Points corresponds to the place where eddy covariance towers are established.

Area Index was measured in May 2019 using a portable canopy analyzer (SunScan, Delta-T Devices, Cambridge, UK). An average of 10 measurements were made on 15 different trees from each experimental site, obtaining values of WF =  $1.89 \pm 0.17 \text{ m}^2 \text{ m}^{-2}$  and WC =  $1.82 \pm 0.13 \text{ m}^2 \text{ m}^{-2}$ . One campaign per month was carried out in 2018 measuring net  $\text{CO}_2$  assimilation ( $A_{\text{leaf}}$ ) and transpiration ( $E_{\text{leaf}}$ ) at leaf level at each treatment, except in November due to instrument failure. Simultaneously, at the ecosystem level, the EC tower provided continuous NEE and  $\text{ET}_{\text{eco}}$  measurements. Time series in two ecosystems were divided into two periods: weed growth before mowing (January-May) and after mowing (June-October). November and December were considered as a transition time when weeds began to grow and proliferate. To highlight the role of the weed cover, we also refer to these periods for the site without cover although this ecosystem was always with bare soil.

## 2.2. Leaf-scale fluxes

Measurements of  $A_{\text{leaf}}$  and  $E_{\text{leaf}}$  were made with a portable photosynthesis system (LI-6800, Li-Cor, Lincoln, NE, USA). In each campaign and treatment, one leaf from each of 10 random olives trees around the EC tower was selected to measure leaf-level gas exchange. Measurements were made at 1.2 m above ground on developed young leaves (soft green leaves at the end of the branch), facing south and  $\pm 2$  h from solar noon on cloudless days. In addition, humidity, light and temperature conditions were controlled for both treatments. Relative humidity was set to 60% to compensate for the loss of humidity of the boundary layer of the leaf,  $\text{CO}_2$  concentration was set at 400 ppm, and photosynthetically active radiation (PAR) to  $1000 \mu\text{mol photons m}^{-2} \text{ s}^{-1}$  since the average light saturation point for olive is  $800\text{--}900 \mu\text{mol m}^{-2} \text{ s}^{-1}$  (Sofa et al., 2009). Throughout the measurements, the temperature inside the chamber was programmed to remain constant for each campaign with the value of the average temperature expected for that day. Measurements were not taken until stabilization occurred: between 2 and 3 min for  $\text{CO}_2$  and 4-5 min for water vapor. Instantaneous water use efficiency at leaf level was calculated as:

$$WUE_{\text{leaf}} = A_{\text{leaf}} / E_{\text{leaf}} \quad (1)$$

Where  $A_{\text{leaf}}$  is the net  $\text{CO}_2$  assimilation rate per unit leaf area ( $\mu\text{mol CO}_2$

$\text{m}^{-2} \text{ s}^{-1}$ ) and  $E_{\text{leaf}}$  is transpiration rate per unit leaf area ( $\text{mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ ).

## 2.3. Ecosystem fluxes

During the study period measurements of NEE and  $\text{ET}_{\text{eco}}$  in the olive grove were measured using EC technique. An EC tower was installed in the centre of each ecosystem, 500 m away from each other to avoid influence from the other treatment (Fig. 1). Instruments were placed 9.3 m high (5.3 m above canopy) to measure densities of  $\text{CO}_2$  and water vapor and wind speed at high frequency (10 Hz). Gas densities were measured by an enclosed-path infrared gas analyser (IRGA, Li-Cor 7200; Lincoln, NE, USA) while the wind speeds in the different vector components were measured by a sonic anemometer (CSAT-3, Campbell Scientific, Logan, UT, USA; hereafter CSI). For more details about EC measurements, see Chamizo et al. (2017).

### 2.3.1. Flux data processing and quality control

Only data which coincided with the time interval of leaf-scale measurement referring to  $\pm 1$  day were analysed. The EddyPro software (v7.0.6) was used to calculate half hourly NEE and  $\text{ET}_{\text{eco}}$ . Spikes, trends, dropouts and discontinuities in eddy covariance data (wind speeds and gas concentration) at 10 Hz were filtered according to Vickers and Mahrt (1997). The time lag between wind speeds and gas concentrations was compensated using covariance maximization. Half hourly means, variances and covariances were calculated applying Reynold's decomposition rules. Double rotation of coordinates and spectral corrections for low frequency (Moncrieff et al., 2006) and high frequency (Fratini et al., 2012) were also applied. Finally, the resulting fluxes were filtered according to the quality control proposed by Mauder et al. (2013), selecting  $qc = 0$  or  $qc = 1$  fluxes. Additional filters were applied to the half hourly fluxes using the methodology described by Chamizo et al. (2017), who also found similar energy balance closures in both ecosystems.

### 2.3.2. Data coverage, missing data and partitioning

EC missing data due to unfavourable meteorological conditions, failures in the instrumentation or quality controls accounted for 27% data losses for measurement in the WF ecosystem (85 % of the data

collected at the leaf level are accompanied by ecosystem data.). Unfortunately, only 38% of leaf-level campaigns in the WC ecosystem are accompanied with concurrent data from the tower. Empirical modelling was used to replace missing data at the ecosystem scale. For the WF ecosystem, only the April 2019 campaign had to be modelled, while for the WC ecosystem, 4 of the 11 campaigns (January 2018, April, October and January 2019) had to be modelled. On the continuous EC database, we used the marginal distribution sampling technique (Reichstein et al., 2005) based on the replacement of missing values using a time window of several adjacent days. After replacing missing data, the semi-empirical model of Reichstein et al. (2005) was applied to split NEE into gross primary production ( $GPP_{eco}$ ) and ecosystem respiration ( $R_{eco}$ ). This method extrapolates to the daytime periods an exponential function of  $R_{eco}$  with temperature using night-time data (assuming that  $GPP_{eco}$  is zero). Missing data replacement and partitioning were done using the online tool REdDyPro R Package (<https://www.bgc-jena.mpg.de/bg/index.php/Services/REddyProcWeb>). Finally, the ecosystem water use efficiency ( $WUE_{eco}$ ) was calculated as:

$$WUE_{eco} = GPP_{eco} / ET_{eco} \quad (2)$$

where  $GPP_{eco}$  is the gross primary production ( $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ ) and  $ET_{eco}$  is evapotranspiration ( $\text{mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ ) of the ecosystem.

## 2.4. Environmental measurements

Complementary environmental measurements were carried out for the two treatments. Temperature and relative humidity of air were measured by a thermohygrometer (HC2S3, Rotronic, AG, Bassersdorf, Switzerland) placed at 5 m height. Soil temperature ( $T_{soil}$ ) was measured in the alleys with two thermocouples at 4 cm below the surface (TCAV, CSI). Volumetric soil water content ( $\theta_v$ ) was measured in the alleys of each treatment with two soil moisture probes installed at a depth of 10 cm in each treatment (CS616, CSI). Incoming and outgoing short-wave and long-wave radiation components were measured by a 4-component radiometer (CNR-4, Kipp and Zonen, Delft, Netherlands) installed at 7 m high and 2 m away from the tower to obtain net radiation and albedo. Incident and reflected PAR were also measured at 7 m using photodiodes (quantum sensor; Li-190, Lincoln, NE, USA). Vapor pressure deficit (VPD) was calculated with data provided by the thermohygrometer. The meteorological data were measured every 30 s, averaged over 30 min, and stored in a data logger (CR3000, CSI).

## 2.5. Nutrient and chlorophyll measurements

The nutrient content was measured in the soil and in the leaves of the olive trees in the two treatments. For soil measurements 6 random samples in the centre of alleys, each composed of 3 subsamples, were taken from 15–25 cm depth in each ecosystem in October. Once dried and ground, the soil organic carbon (SOC) content was measured by a modified wet oxidation method (Mingorance et al., 2007) and determined with a spectrophotometer (Spectronic Helios Alpha 9423 UVA 1002E, Thermo Fisher Scientific, Waltham, MA, USA). For other nutrients, samples were sent to a laboratory (CSR Laboratorio, Ubeda, Jaen). Nitrogen was determined using Kjeldahl method, phosphorus was determined by ultraviolet-visible spectroscopy and potassium, calcium, magnesium and sodium were determined by atomic absorption spectroscopy. Also, one leaf per cardinal direction in 25 random olive trees was collected from each ecosystem in January. Once dried and ground, the sample was subdivided into 6 subsamples for subsequent analysis. Nitrogen was determined by the Spectrophotometric Krom Method (1980) and other elements were obtained by plasma emission spectrometry after digesting the solid samples with acid.

From July 2018 to December 2019, 14 apical leaves of different trees from each treatment were collected monthly to measure the chlorophyll content. Total chlorophyll (Chl) was extracted in methanol and

centrifuged at  $5000 \times g$  for 5 min. Thereafter, the absorbance of the supernatant was measured at 664, 648, and 470 nm. The chlorophyll a (Chl a), chlorophyll b (Chl b), and carotenes were estimated by using the equation of Lichtenthaler described in Wellburn (1994).

## 2.6. Statistical analysis

Statistical tests of two independent samples (t-student) were performed on these subsets to identify significant differences in fluxes between treatments and periods. The Shapiro-Wilk test determined normality of variables. To ensure normality, box-cox transformations ( $A_{leaf}$ ,  $WUE_{leaf}$ ,  $R_{eco}$ ,  $GPP_{eco}$ ,  $WUE_{eco}$ ) and transformations based on logarithms and square roots ( $E_{leaf}$ ,  $ET_{eco}$ ,  $T_{air}$ , VPD,  $\theta_v$ ) were applied. After that, homoscedasticity was evaluated before running the t-tests.

Due to the small sample size, non-parametric Mann-Whitney tests were used to compare nutritional status between treatments ( $n = 6$ ) and the averages fluxes in each individual campaign ( $n = 10$  and  $n = 9$  for leaf and ecosystem fluxes). The graphs and statistical analyses were carried out using the software Matlab (version R2020a).

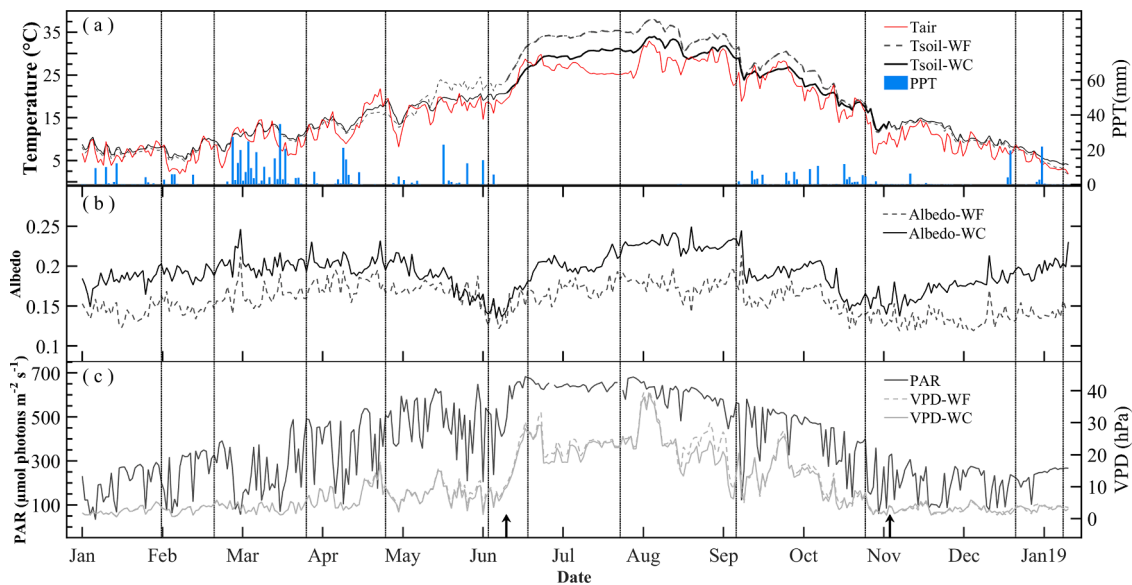
## 3. Results

### 3.1. Meteorological conditions and macronutrients

Meteorological variables showed large seasonal variation in the olive grove (Fig. 2). Winter months (January–February) presented the lowest daily values of air temperatures ( $7 \pm 3^\circ\text{C}$ ), PAR ( $228 \pm 99 \mu\text{mol photons m}^{-2} \text{ s}^{-1}$ , minimum  $< 40 \mu\text{mol photons m}^{-2} \text{ s}^{-1}$  on cloudy days) and VPD ( $3 \pm 1 \text{ hPa}$ ). The annual precipitation was 536 mm with precipitation events mainly in autumn and especially in spring when numerous episodes of rain were quantified with up to 6 events of more than 20 mm  $\text{day}^{-1}$ , with maximum of 35 mm  $\text{day}^{-1}$ . On average the albedo of the WC ecosystem was 23% higher (t-test;  $p < 0.001$ ) than that of WF. Albedo ranged between 0.13 and 0.15 for the weed-free olive ecosystem while with weed cover the albedo was more fluctuating with values between 0.12 and 0.21 (Fig. 2b). Only several weeks before mowing did albedos become similar. After the harvest the differences were established again. The reflected PAR in WF was 60% greater than in WC during weed growth, while it was only 25% greater after weed mowing.

In July–August, high average daily air temperatures ( $27 \pm 2^\circ\text{C}$ ) coincided with the absence of precipitation, high average daily PAR ( $635 \pm 80 \mu\text{mol photons m}^{-2} \text{ s}^{-1}$ , reaching 2080  $\mu\text{mol photons m}^{-2} \text{ s}^{-1}$  at solar noon) and high daily VPD ( $24 \pm 5 \text{ hPa}$ ), typical of the Mediterranean summer. For the weed-covered olive ecosystem, this dry and warm period coincided with the mowed period. Significant differences in soil temperatures and VPD between treatments were observed during this period (t-test;  $p < 0.001$ ). On average, soil temperatures and VPD were  $5^\circ\text{C}$  and 2 hPa lower in the olives with mowed weeds versus the olive ecosystem with bare soil.

Additionally, the two treatments presented differences in field capacity (WC =  $46.7 \pm 0.9 \%$ ; WF =  $30.1 \pm 3.5 \%$ ) and in soil and leaf nutritional status (Table 1). The soil of the weed-covered ecosystem had higher nitrogen, potassium, calcium, magnesium and calcium, with an 82% exchangeable cation content. There was an excess of K and Mg in the weed-covered soil. In addition, the organic carbon content in this soil was 68% higher. Although the values of macronutrients were higher in the leaves of the weed-free ecosystem, no significant differences were detected between treatments in the foliar analysis. However, there were differences in some micronutrients. Lower concentrations of zinc and higher concentrations of boron and molybdenum were found in the leaves of the WF ecosystem. Finally, no differences were detected in the concentrations of chlorophyll a ( $Cl a_{wc} = 0.065 \pm 0.029 \text{ mg g}^{-1}$ ;  $Cl a_{wf} = 0.067 \pm 0.027 \text{ mg g}^{-1}$ ), chlorophyll b ( $Cl b_{wc} = 0.040 \pm 0.007 \text{ mg g}^{-1}$ ;  $Cl b_{wf} = 0.040 \pm 0.007 \text{ mg g}^{-1}$ ) and carotenes ( $Carotenes_{wc} = 41.5 \pm 12.2 \text{ mg g}^{-1}$ ;  $Carotenes_{wf} = 40.4 \pm 13.3 \text{ mg g}^{-1}$ ).



**Fig. 2.** Daily averages for each experimental ecosystem (weed cover (WC) and weed free (WF)) of (a) air temperature, soil temperatures in the weed covered and weed free soil and cumulative daily precipitation (PPT); (b) albedo; (c) incident photosynthetic active radiation (PAR) and vapor pressure deficit (VPD). Since no differences between ecosystems were observed for air temperature and PAR, only the data for WF were shown for clarity. Vertical dashed lines indicate leaf gas exchange measurement campaigns. Arrows indicate the moment when weeds were mowed (left) and the period in which the weeds were already consolidated and growing (right) in the WC ecosystem.

**Table 1**  
Summary of chemical parameters in soils and leaves for both treatments.

Soil	N (g/kg)	P (mg/kg)	K (mg/kg)	Ca (mg/kg)	Mg (mg/kg)	S	Na (mg/kg)	SOC (%)
Weed Free	0.7 ± 0.1**	6.0 ± 1.7	210 ± 10**	3.1 ± 0.4**	340 ± 60**	–	69 ± 7**	0.6 ± 0.1**1.0 ± 0.2
Weed Covered	0.9 ± 0.1	8.3 ± 4.1	450 ± 50	5.2 ± 0.2	750 ± 30	–	136 ± 11	–
<b>Macronutrients in leaves</b>	<b>N (g/kg)</b>	<b>P (g/kg)</b>	<b>K (g/kg)</b>	<b>Ca (g/kg)</b>	<b>Mg (g/kg)</b>	<b>S (g/kg)</b>	–	–
Weed Free	17.1 ± 3.3	1.43 ± 0.05	7.3 ± 0.9	21.6 ± 1.4	2.36 ± 0.12	1.99 ± 0.06	–	–
Weed Covered	15.3 ± 1.7	1.38 ± 0.05	7.1 ± 0.4	21.3 ± 0.5	2.19 ± 0.16	2.00 ± 0.09	–	–
<b>Micronutrients in leaves</b>	<b>Fe (mg/kg)</b>	<b>Cu (mg/kg)</b>	<b>Zn (mg/kg)</b>	<b>B (mg/kg)</b>	<b>Mn (mg/kg)</b>	<b>Mo (mg/kg)</b>	<b>Na (mg/kg)</b>	–
Weed Free	250 ± 40	190 ± 40	29.8 ± 3.7	42 ± 11**	84.5 ± 2.9	0.33 ± 0.09	76 ± 10	–
Weed Covered	320 ± 80	230 ± 20	40.0 ± 3.8	27 ± 2	81.2 ± 9.1	0.21 ± 0.05	90 ± 15	–

Note. Soil Organic Carbon (SOC) and carbon. Weights (g/kg) are per kilogram of dry weight. Stars indicate significant difference between treatments (Mann-Whitney test)

\*p < 0.05; n = 6

\*\*p < 0.01; n = 6

### 3.2. Carbon dioxide and water vapor fluxes

#### 3.2.1. Leaf level

During the entire study period,  $A_{\text{leaf}}$  was significantly different between treatments (t-test;  $p < 0.001$ ;  $n = 110$ ). In the WF olive ecosystem,  $A_{\text{leaf}}$  was 24% higher than in the WC ecosystem with mean values of  $A_{\text{leaf-WF}} = 9.3 \pm 4.4 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ , and  $A_{\text{leaf-WC}} = 7.5 \pm 4.8 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ . Mean values of  $E_{\text{leaf}}$  and  $WUE_{\text{leaf}}$  for the whole period were  $E_{\text{leaf-WF}} = 1.2 \pm 1.1 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ ;  $E_{\text{leaf-WC}} = 1.0 \pm 0.9 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$  (20% higher in WF ecosystem; t-test;  $p = 0.791$ ;  $n = 110$ ) and  $WUE_{\text{leaf}}$  was 16% higher in WF ecosystem; (t-test;  $p = 0.051$ ;  $n = 110$ ).

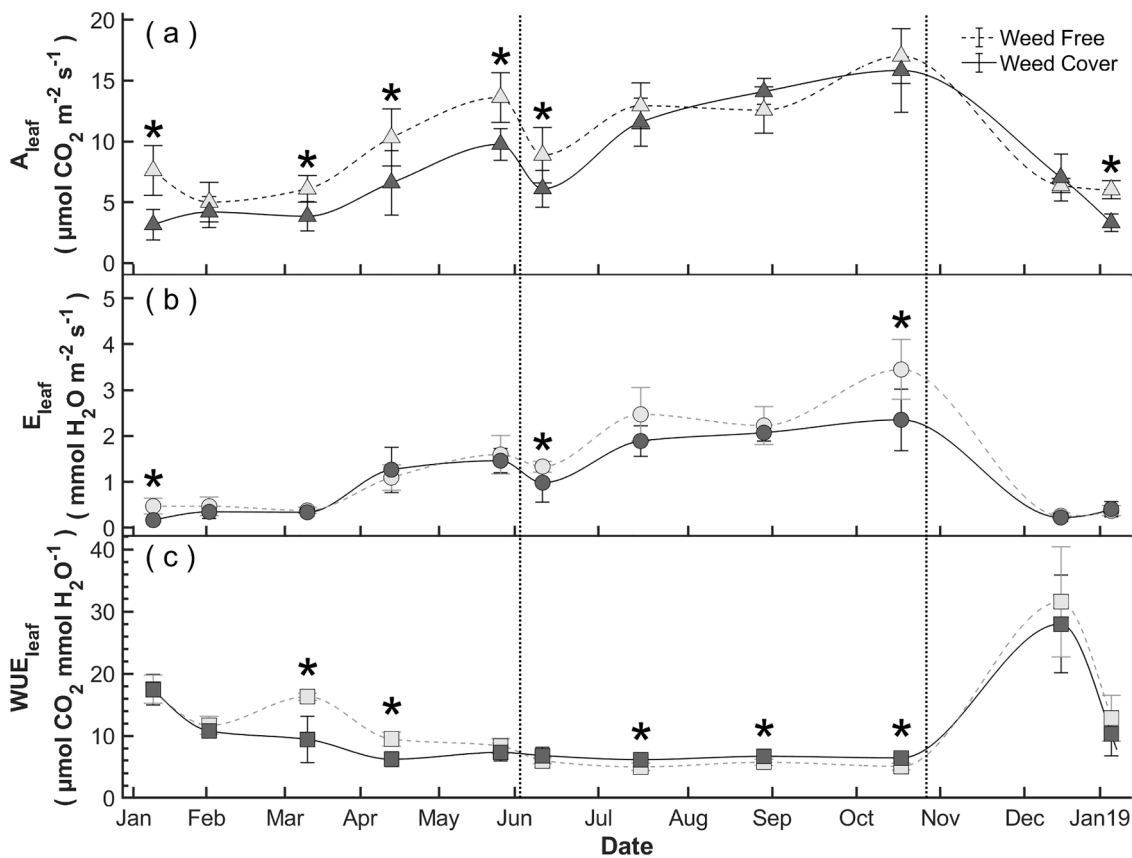
Regarding temporal variability, we observed seasonal variations with asynchronous patterns between photosynthesis and  $WUE_{\text{leaf}}$  throughout the year (Fig. 3). Specifically, we observed higher values of  $A_{\text{leaf}}$  and  $E_{\text{leaf}}$  and low values of  $WUE_{\text{leaf}}$  on high VPD days and low values of  $A_{\text{leaf}}$  and  $E_{\text{leaf}}$  and higher values of  $WUE_{\text{leaf}}$  on low VPD days.  $A_{\text{leaf-WF}}$  data ranged between a minimum of  $5.0 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$  in February and a maximum of  $17.0 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$  in October (Fig. 3a). On the other hand,  $A_{\text{leaf-WC}}$  ranged between a minimum of  $3.2 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$  in January and  $15.8 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$  also in October. April was the month that, on average, had the most fluctuating values, with standard deviation of  $5.1 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$  while March had the least,

with a standard deviation of  $2.1 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ . Significant differences between treatments on  $A_{\text{leaf}}$  were found for winter and spring campaigns (Mann-Whitney test;  $p < 0.001$ ;  $n = 10$ ). These differences were found when weeds were present and persisted into June (7 days after mowing), but not through the summer.

Transpiration differed significantly (Mann-Whitney test;  $p < 0.001$ ;  $n = 10$ ) between treatments for January, June and October with more transpiration in the leaves of weed-free olive ecosystem (Fig. 3b). In fact,  $E_{\text{leaf-WF}}$  reached a maximum of  $3.4 \pm 0.9 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$  in October, exceeding the maximum in the WC ecosystem ( $E_{\text{leaf-WC}} = 2.4 \pm 0.9 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ ) in the same month. Double and triple values of  $WUE_{\text{leaf}}$  were observed in winter in comparison with the annual average (Fig. 3c). However, variations in  $WUE_{\text{leaf}}$  were lower in the leaves of the WC olive ecosystem ( $WUE_{\text{leaf-WC}} = [5.6 - 28.0] \mu\text{mol CO}_2 \text{ mmol}^{-1} \text{ H}_2\text{O}$ ) than in WF ( $WUE_{\text{leaf-WF}} = [4.9 - 31.6] \mu\text{mol CO}_2 \text{ mmol}^{-1} \text{ H}_2\text{O}$ ). For March and April  $WUE_{\text{leaf-WF}} > WUE_{\text{leaf-WC}}$  while for July, August and October  $WUE_{\text{leaf-WF}} < WUE_{\text{leaf-WC}}$  (Mann-Whitney test;  $p < 0.001$ ;  $n = 10$ ).

#### 3.2.2. Ecosystem level

During the study period,  $GPP_{\text{eco}}$  and  $ET_{\text{eco}}$  were significantly different between treatments (t-test;  $p < 0.001$ ;  $n = 99$ ; values at the



**Fig. 3.** Average and standard deviation of leaf-level carbon and water fluxes for the two experimental sites. (a) net  $\text{CO}_2$  assimilation rate ( $A_{\text{leaf}}$ ), (b) transpiration ( $E_{\text{leaf}}$ ) and (c) water use efficiency ( $\text{WUE}_{\text{leaf}}$ ). Dashed (weed free) and continuous (weed cover) lines were interpolated values using a piecewise cubic polynomial. Stars indicate significant differences between treatments (Mann-Whitney test;  $p < 0.001$ ;  $n = 10$ ). Vertical dashed lines indicate the moment when weeds were mowed (left) and the period in which the weeds were already consolidated and growing (right) in the WC ecosystem. Sampling Days (as Day Of Year; DOY): 31, 51, 86, 116, 155, 170, 205, 249, 264, 298, 355, 8 (2019).

same time of day when leaf data were taken).  $\text{GPP}_{\text{eco}}$  for the weed-free olive ecosystem was 32% lower than in the WC ecosystem with values of  $\text{GPP}_{\text{eco-WF}} = 8.2 \pm 2.3 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$  and  $\text{GPP}_{\text{eco-WC}} = 12.12 \pm 4.5 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ . Similarly,  $\text{ET}_{\text{eco-WF}} = 2.2 \pm 1.1 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$  was 22% lower than  $\text{ET}_{\text{eco}}$  in WC ecosystem, which had  $\text{ET}_{\text{eco-WC}} = 2.8 \pm 1.1 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ . However, no significant differences were observed in  $\text{WUE}_{\text{eco}}$ .

Ecosystem fluxes are strongly seasonal with high values of  $\text{GPP}_{\text{eco}}$  and  $\text{ET}_{\text{eco}}$  in spring, low values of  $\text{GPP}_{\text{eco}}$  and  $\text{WUE}_{\text{eco}}$  in summer and high values of  $\text{WUE}_{\text{eco}}$  on cold days (Fig. 4). Also, this variability in carbon and water fluxes is more pronounced for the WC than the WF ecosystem. As the herbaceous plants grew, the differences in  $\text{GPP}_{\text{eco}}$  and  $\text{ET}_{\text{eco}}$  between treatments were accentuated (Fig. 4a and b).  $\text{GPP}_{\text{eco-WF}}$  ranged between  $5.8 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$  in July and  $11.6 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$  in April. On the other hand,  $\text{GPP}_{\text{eco-WC}}$  ranged between  $5.4 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$  in June and  $18.3 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$  in March.  $\text{ET}_{\text{eco-WF}}$  ranged between 0.8 and  $3.6 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$  in December and June respectively while for  $\text{ET}_{\text{eco-WC}}$  ranged between 1.6 and  $4.4 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$  in August and May respectively. Higher values of  $\text{WUE}_{\text{eco}}$  were observed during winter compared to the annual average for both treatments (Fig. 4c).  $\text{WUE}_{\text{eco-WF}}$  ranged between 2.4 and  $11.2 \mu\text{mol CO}_2 \text{ mmol}^{-1} \text{ H}_2\text{O}$  in June-July and December while  $\text{WUE}_{\text{eco-WC}}$  between 2.0 and  $7.1 \mu\text{mol CO}_2 \text{ mmol}^{-1} \text{ H}_2\text{O}$ . Significant differences between treatments were found in 9, 7 and 4 out of 11 campaigns for  $\text{GPP}_{\text{eco}}$ ,  $\text{ET}_{\text{eco}}$  and  $\text{WUE}_{\text{eco}}$  respectively (Mann-Whitney test;  $p < 0.001$ ;  $n = 9$ ).

### 3.3. Weed effects on olive leaf fluxes

With the database split between weed growing and mowed periods

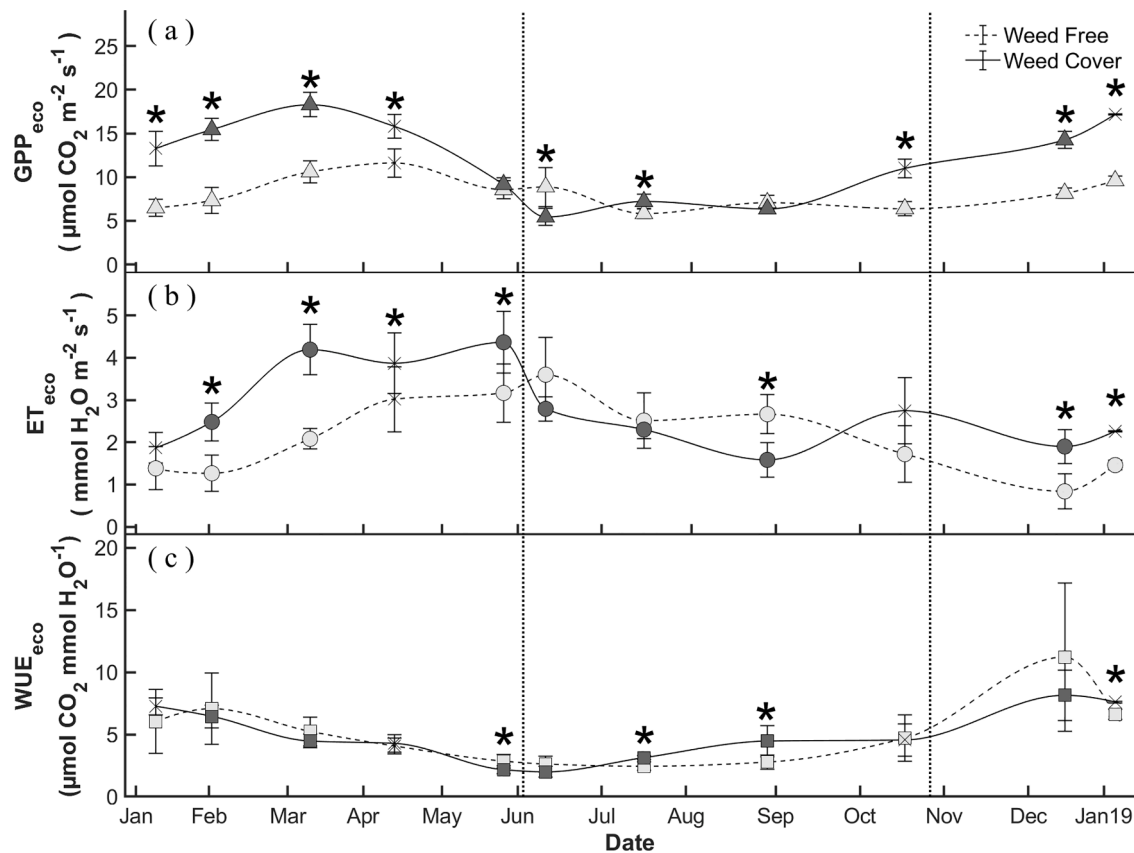
we can see that observed differences between treatments on  $A_{\text{leaf}}$  are produced when weeds were growing (Fig. 5a).  $A_{\text{leaf-WC}}$  is 33% lower than  $A_{\text{leaf-WF}}$  (t-test;  $p < 0.001$ ;  $n=60$ ) during weed growth ( $A_{\text{leaf-WC}} = 5.1 \pm 3.1 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ ;  $A_{\text{leaf-WF}} = 7.6 \pm 3.7 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ ) and it is similar after mowing. At the same time, gross primary production ( $\text{GPP}_{\text{eco}}$ ) by the WF olive ecosystem was 39% lower than  $\text{GPP}_{\text{eco-WC}}$  when weeds were growing but after mowing  $\text{GPP}_{\text{eco}}$  was similar in both ecosystems (Fig. 5d).

Similarly, WC  $\text{ET}_{\text{eco}}$  was 43% more than WF during weed growth only (Fig. 5e) (t-test;  $p < 0.001$ ;  $n=54$ ). Unlike carbon fluxes,  $E_{\text{leaf}}$  was similar for both treatments during this period (Fig. 5b). After mowing, there was a significant difference in the water fluxes at the leaf level ( $E_{\text{leaf-WC}} = 1.8 \pm 0.8 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$  and  $E_{\text{leaf-WF}} = 2.4 \pm 1 \text{ mmol H}_2\text{O m}^{-2} \text{ s}^{-1}$ ) but not at the ecosystem level. Finally, although no differences were observed between periods at the ecosystem scale in  $\text{WUE}$ , significant differences were observed at the leaf level (Fig. 5c and f), with more  $\text{WUE}$  in the leaves without weed cover during the period before mowing ( $\text{WUE}_{\text{leaf-WC}} = 9.8 \pm 4.8 \mu\text{mol CO}_2 \text{ mmol}^{-1} \text{ H}_2\text{O}$  and  $\text{WUE}_{\text{leaf-WF}} = 12.6 \pm 4.2 \mu\text{mol CO}_2 \text{ mmol}^{-1} \text{ H}_2\text{O}$ ) and more  $\text{WUE}$  in the leaves with weed cover in the mowed period ( $\text{WUE}_{\text{leaf-WC}} = 6.7 \pm 1.2 \mu\text{mol CO}_2 \text{ mmol}^{-1} \text{ H}_2\text{O}$  and  $\text{WUE}_{\text{leaf-WF}} = 5.5 \pm 0.8 \mu\text{mol CO}_2 \text{ mmol}^{-1} \text{ H}_2\text{O}$ ).

## 4. Discussion

### 4.1. Carbon fluxes

Higher SOC were found in the alleys of the WC olive ecosystem (Table 1) supporting the higher uptake of  $\text{CO}_2$  in the WC ecosystem



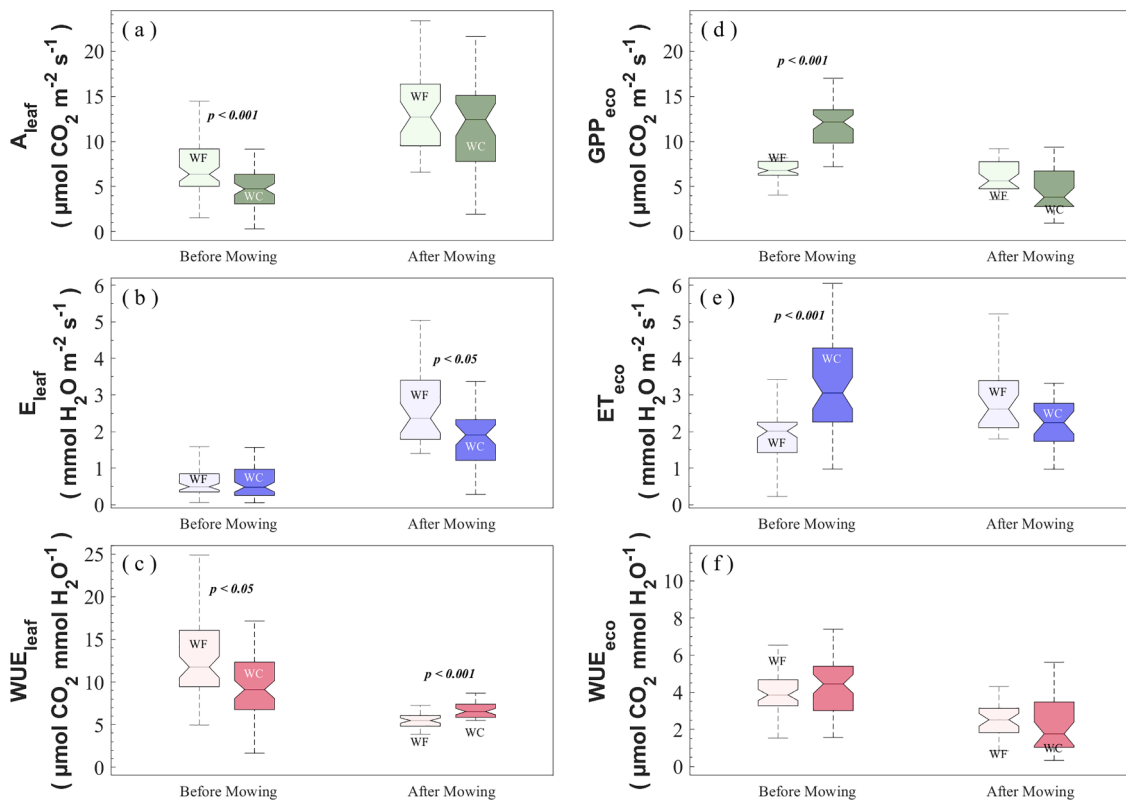
**Fig. 4.** Average and standard deviation of ecosystem carbon and water fluxes for the two experimental sites at the same moment when leaf data were taken. (a) gross primary production ( $GPP_{eco}$ ), (b) evapotranspiration ( $ET_{eco}$ ) and (c) ecosystem water use efficiency ( $WUE_{eco}$ ). Dashed (weed free) and continuous (weed cover) lines were interpolated values using a piecewise cubic polynomial. Modelled data are represented with an “x”. Asterisks indicate significant difference between treatments (Mann-Whitney test;  $p < 0.001$ ;  $n = 9$ ). Vertical dashed lines indicate the moment when weeds were mowed (left) and the period in which the weeds were already consolidated and growing (right) in the WC ecosystem. The standard deviation represents the variability in ecosystem behaviour due to varying environmental conditions since is higher than erratic nature of eddy fluxes.

found in previous studies. In fact, during the weed growth period  $GPP_{eco}$  (at the same moment when leaf data were taken) was 65% higher in WC than in WF. However, in this study we show that these differences in carbon assimilation at the ecosystem level correspond not only to an increase in carbon removal by weeds but also a decrease in carbon sequestration by the main crop. The olive leaves of WC assimilated 33% less  $CO_2$  than those of WF when weeds were established, which can be detrimental to crop growth. In other studies that compare olive groves with and without cover weed (Chamizo et al., 2017), equal carbon assimilation in olive trees has been assumed. However, in a study we show that the photosynthetic capacity of olive trees is affected by weeds revealing the importance of studying fluxes in the various components of orchards.

Seasonal trends in leaf-scale carbon fluxes were similar for both ecosystems (Fig. 3a), while at ecosystem scale these patterns were more accentuated in the WC ecosystem (Fig. 4a), corroborating the great influence of the weeds on carbon sequestration. Chamizo et al. (2017) showed that the weed growth coincides not only with high rates of carbon sequestration by the ecosystem but also with the highest soil  $CO_2$  efflux. In fact, in our study net  $CO_2$  uptake was 42% higher respectively in WC than in WF while GPP was 65% higher. These differences are due to a greater increase in  $CO_2$  uptake induced by weed cover with respect to a slight increase in soil respiration. Although trunks and branches contribute a small fraction of olive tree respiration, leaves are the main contributors to above-ground plant respiration (Pérez-Priego et al., 2014) and the autotrophic respiration of trees at the leaf and ground level could also change with the establishment of the weed cover. Nonetheless we can deduce that herbaceous plants contribute

substantially to the respiration of the whole agroecosystem, but this carbon emission does not even represent  $\sim 1/3$  of the carbon that is assimilated by weeds. Therefore, although continuous monitoring of soil or subcanopy  $CO_2$  fluxes under the two treatments could help to quantify the role of weed cover in NEE or  $GPP_{eco}$ , we can say that weeds enhanced olive groves as carbon sinks despite the olive trees sequestering less carbon and the soil emitting more  $CO_2$ .

One of the most remarkable results of this study was that  $A_{leaf}$  was not different between treatments after mowing. This fact is supported by similar values of  $GPP_{eco}$  in both treatments after mowing (when all the carbon assimilation comes from the olive trees). In contrast, net  $CO_2$  uptake was 20% lower in WC after mowing, due to increased soil respiration (as the weed roots have died and are mineralized) and the decomposition of the surface weed biomass. However, it took weeks for  $A_{leaf}$  to progressively equalize across treatments and, according to Fig. 3a we can say that differences in  $A_{leaf}$  persisted beyond 10 days after mowing (June), which could have repercussions for fruit growth. In June the fruit begins to develop, so if the olive tree is assimilating less carbon it could translate to less yield. Similarly, the return to differences in  $A_{leaf}$  between treatments is manifested in the last winter season (January) approximately 10 weeks after weeds started growing in WC (the moment when  $GPP_{eco-WC} \gg GPP_{eco-WF}$ ) which seems to indicate that the presence of weeds only modifies  $A_{leaf}$  from a certain stage of herbaceous plant growth. This may have repercussions for the management of the olive grove: on the one hand, sooner mowing could allow olive leaves to recover from the effects of competition with weeds, which could imply improvements in crop yield. On the other hand, maximizing the period in which there is no differences in  $A_{leaf}$  (no effect on the crop)



**Fig. 5.** Box-plots of carbon and water fluxes for periods before (January–May) and after weed mowing (June–October) in the two experimental sites. (a–c) leaf-scale values of (a) net  $\text{CO}_2$  assimilation rate ( $A_{\text{leaf}}$ ), (b) transpiration ( $E_{\text{leaf}}$ ), and (c) water use efficiency ( $\text{WUE}_{\text{leaf}}$ ); and (d–f) ecosystem-scale values of (d) gross primary production ( $\text{GPP}_{\text{eco}}$ ), (e) evapotranspiration ( $\text{ET}_{\text{eco}}$ ) and (f) water use efficiency ( $\text{WUE}_{\text{eco}}$ ). WF: weed free ecosystem. WC: weed cover ecosystem. Whiskers are  $\pm 2.7\sigma$ . Significant differences are represented by corresponding p value. The notch ends correspond to  $q_2 - 1.57 (q_3 - q_1) / \sqrt{n}$  and  $q_2 + 1.57 (q_3 - q_1) / \sqrt{n}$ . Where  $q_2$  is the median,  $q_1$  and  $q_3$  are the 25th percentiles and 75, respectively.

but there is more  $\text{GPP}_{\text{eco}}$  may be a suitable approach to find a balance between agronomic and ecological benefits. Although studies that quantify  $A_{\text{leaf}}$  throughout the whole year are scarce, we found that the range of average values in each campaign [ $3\text{--}17 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ ] is similar to another irrigated olive grove where similar methodologies were applied throughout a year (Bedbabis et al., 2017). In our olive grove, both treatments showed similar temporal patterns over the year. Increased  $A_{\text{leaf}}$  was observed during the spring months (from March to May), followed by a sharp decline at the beginning of summer (June), a progressive increase until mid-autumn (maximum in October), and a decrease again during the winter months (December–January, Fig. 3). This temporal pattern is also described in sub-annual-scale studies for both irrigated (Proietti and Famiani, 2002) and non-irrigated (Moriana et al., 2002; Hagidimitriou and Pontikis, 2005; Proietti et al., 2012) olive groves. Hydraulic stress on stomata due to high VPD and low soil water content could be the factors which cause the low values of  $A_{\text{leaf}}$  in summer. High values of  $A_{\text{leaf}}$  in October ( $15\text{--}17 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$ ) have been observed in other studies (Moriana et al., 2002; Villalobos et al., 2012; López-Bernal et al., 2015) and may be presumably due to the better ambient conditions (VPD = 1.2 kPa;  $T_{\text{air}} = 23^\circ\text{C}$ ); in addition to the availability of water in the alleys due to precipitation.

However, the comparison between different studies is only reliable with the same temporal methodology. The patterns described could be affected by differences in the time of day or the year in which data were collected. The leaf fluxes in olive trees have an asymmetric daily pattern in the summer. In the warmer months, the maximum photosynthetic rates in olive groves are always reached before noon (Angelopoulos et al., 1996; Moriana et al., 2002; Testi et al., 2008) since VPD is lower in the morning, the stomatal conductance is higher and consequently so too are  $\text{WUE}_{\text{leaf}}$  and  $A_{\text{leaf}}$ . On the other hand, the olive tree is an alternate fruit-bearing species, and it is characterized by its ability to alternate

years of high and low yields. For instance, Bedbabis et al. (2017) detected a greater decline in  $A_{\text{leaf}}$  in June in a high fruit load year than in a low fruit load. This reduction in assimilation seems to coincide with the phase of suspension of fruit growth and pit hardening (Rapoport et al., 2013). Another point to consider is the within-tree variability of leaf-scale fluxes because of the leaves being exposed to very different light intensities within the canopy. For example, assimilation rates have been observed to decrease significantly and progressively from the highest parts of the tree to the lowest (Larbi et al., 2015) or to vary with leaf age (Hagidimitriou and Pontikis, 2005; Proietti et al., 2012). Also, shaded leaves and unshaded leaves have different rates of photosynthesis in the olive tree even applying light saturation (Gregorio et al., 2007; Sofo et al., 2009). In order to compare between treatments, the orientation and the measurement height were kept constant in this study, but there may be other variables that influence gas exchanges that are not being considered, such as leaf angle or age.

#### 4.2. Water vapor fluxes and water use efficiency

Differences in  $E_{\text{leaf}}$  between treatments were manifested only in summer and after mowing. Since the field capacity was higher in the WC ecosystem and the olive trees received the same irrigation in both treatments, higher transpiration in summer would be expected due to more water available to the olive tree. However, we found 25% less olive transpiration in WC treatment with a mowed layer of weeds. Mowed weeds reduced soil temperature and VPD, conditions that may reduce the need for transpiration for cooling even if there is more water available to transpire. Regarding  $\text{ET}_{\text{eco}}$  after mowing; although the alleys are dry and  $\text{ET}_{\text{eco}}$  is mainly due to  $E_{\text{leaf}}$  by the olive trees (Testi et al., 2004), no clear significant differences were shown between treatments (Fig. 4b). Because the alleys are dry after mowing, irrigation (same



amount and frequency in both treatments) is still provided in this period, so soil evaporation could be affecting the differences in  $ET_{eco}$  between an ecosystem with mowed weed and one with bare soil. The evaporative fraction has not been determined but it could also change between treatments due to a reduction by a layer of mowed weeds. Daily averages in  $ET_{eco}$  showed that only during the 15 days after mowing there were decreases in the  $ET_{eco}$  of the WC with respect to the WF ecosystem (data not shown). Therefore, we can cautiously affirm that  $E_{leaf}$  of the olive trees decreased with weed mowing, but we cannot affirm that  $ET_{eco}$  decreased (if  $ET_{eco}$  was lower in the ecosystem with mowed weed no significant differences were detected). On the other hand, during the growing period no differences were observed in transpiration of the olive trees (Fig. 3b), but there were notable differences in  $ET_{eco}$  between treatments (Fig. 4b). Since more than 95% of the ET in developed weeds corresponds to transpiration (Rothfuss et al., 2010) we can infer that the transpiration of the herbaceous plants is causing the difference in  $ET_{eco}$  between treatments.

There are few studies that quantify the seasonal variability of  $E_{leaf}$  and  $WUE_{leaf}$  by direct measurement with a portable gas analyser in an olive grove. Water vapor fluxes were measured normally in olive groves with other techniques such as: sap flow sensors (López-Bernal et al., 2015; Ayyoub et al., 2017), chambers (Villalobos et al., 2012) or semi-empirical models (Yunusa et al., 2008; Torres-Ruiz et al., 2012). In our study we can see that the irrigated period [DOY ~ 116, DOY ~ 298], coincides with higher rates of  $E_{leaf}$ , with maxima in October (Fig. 3b) which coincided with the maximum values of  $A_{leaf}$ . In other studies, by contrast, transpiration increased over the course of the summer (Proietti et al., 2012; Villalobos et al., 2012), but the peak in transpiration was detected in August when there were higher temperatures (and VPD) and not in October. The maximum in transpiration in the middle of summer was also found by Yunusa et al. (2008). López-Bernal et al. (2015) found that tree-scale transpiration (from sap flow measurements) even decreased in two of their three years of study over the course of the summer. October data were taken at a time after some rains after which irrigation continued. It could be that the  $E_{leaf}$  is more variable and depends on the amount and management of the irrigation applied, as well as the meteorological conditions of the year.

Although  $E_{leaf}$  increased in the irrigation period (from April to October),  $WUE_{leaf}$  was the lowest during this period in both treatments. On the contrary,  $WUE_{leaf}$  was greatest during colder months when VPD was minimum (January, February and December). A sharp increase was observed in late autumn - early winter. Such changes were detected in our study both at the leaf and ecosystem scales (Figs. 3c and 4c) and have also been observed in olive groves (Testi et al., 2008; Villalobos et al., 2012). Although the changes in  $WUE_{leaf}$  were dependent on VPD (Table 3), the high values found in winter (VPD < 5 hPa) must be related to variables that are not meteorological (Testi et al., 2006). In such a way, an approach based on studying the phenological moments in olive trees could be more appropriate to study unusual  $WUE$  values.

#### 4.3. Effect of weed cover on olive leaf fluxes

A lower carbon assimilation in the leaves only when the weed cover is developed suggest an effect of weeds on olive-tree behaviour. After mowing,  $A_{leaf}$  showed no difference between treatments and  $WUE_{leaf}$  was higher in WC ecosystem. These facts show a competition between the weed cover and the tree. In other Mediterranean crops, like grapevines, weed presence causes a conspicuous reduction in  $A_{leaf}$  (Pou et al., 2011). With the data obtained in this study, it is difficult to evaluate the resource for which competition is taking place. However, the presence of weed can change some parameters in the environment which in turn affect carbon fluxes from the leaf.

The results of Fig. 2 reaffirmed that the presence of weed cover reduced soil temperature fluctuations as with laboratory results regarding mulching (Montenegro et al., 2013). Differences were detected not only in the soil variables but also in the atmosphere, especially in

the VPD, reflecting different meteorological states between treatments. The weed cover had the potential to reduce the VPD of the environment in summer (Fig. 1a). It has long been known that VPD is a fundamental variable on which gas exchanges depend since olive trees can close their stomata in high VPD to avoid excessive loss of water (Fernández et al., 1999). Since VPD was lower in the WC ecosystem, especially with a layer of mowed weed, the presence of weed cover may have increased the efficiency in the use of water by olive trees. In fact, the small difference in VPD after mowing are consistent with significantly high  $WUE_{leaf}$  in the WC ecosystem after mowing.

Additionally, other meteorological variables could influence  $A_{leaf}$  and  $E_{leaf}$ . Weed cover and residues can increase albedo and thus indirectly increase carbon assimilation (Carrer et al., 2018) by an increase in diffuse radiation reflected from the ground and received on the underside of olive leaves. In our study, the albedo was 23% higher in the WC when weeds were present (17% on sampling days), while  $A_{leaf}$  was lower compared to WF. On the other hand, the reflected PAR in WF was 60% greater than in WC when weeds were present, but only 25% greater after weeds were mowed. Therefore, wavelengths outside the PAR must be energetically dominant in determining the difference in albedo. This increase in reflected PAR could also have contributed to the differences in the assimilation of the olive tree. Bare soil reflects longer PAR wavelengths than weeds to the lower canopy of olive trees, which in turn could be related to differences in  $A_{leaf}$  with height (Larbi et al., 2015). This promotes more assimilation by weed-free olives.

Although differences in SOC could mean higher water storage as shown by field capacity, differences in SOC could not explain differences in leaf-level fluxes since once mowed the fluxes equalized in both ecosystems but SOC remained the same. In addition, the chemical quality of the soils in the WC olive ecosystem was better in general, but higher amounts of macronutrients in soil were not reflected in the leaf nutrients of this ecosystem (Table 1). In our study we observed that the treatment with less macronutrients in soil was the treatment with more macronutrients in the leaves, while the nutrient input (via ferti-irrigation) was the same in both treatments. Also, the fertilization method was done by irrigation at the tree base, while the olive roots can extend underground for several meters horizontally. The tree and herbaceous plants compete for soil nutrients beyond the irrigation bulb. This fact supports the idea that the neighbouring herbaceous plants could assimilate the nutrients that are finally partially integrated into the soil instead of being assimilated by the crop.

Another likely competitive factor is water. With the abundance of light, the availability of water tends to stand out as the main limiting factor for photosynthesis in Mediterranean environments. As suggested by the higher evapotranspiration in the WC ecosystem when weeds were growing, the need for water was greater in the presence of herbaceous plants. Since the amount of irrigation was the same in both treatments, this could reflect a water deficit for olives in the WC ecosystem. Nevertheless, notice that transpiration in olive leaves was the same in both treatments when weeds were present. This seems not to reflect competition for water. Soil water content under canopy would be a good indicator to observe the availability of water for the olive tree. Unfortunately, our soil water content sensors were in the alleys and not at the base of the tree. Therefore, more study is needed to monitor the hydric status of the leaves and the soil to help us clarify whether the difference in fluxes is linked to water availability.

Additionally, the presence of weeds was manifested in the olive yield. In January 2019, the olives from 100 olive trees were harvested in each treatment and weighed by means of a bascule. A 31% difference was observed between treatments, with 90.0 kg per olive tree in the WF ecosystem and 62.4 kg per olive tree in the WC ecosystem. This difference in production was also detected in other weed-covered olive groves managed with Sheep-walk (Ferreira et al., 2013). These yield results could fuel the debate regarding the best time to mow. Mowing coincides with the fruiting period and advancing it could improve carbon assimilation with less water use, which might reduce the differences in yield.

## 5. Conclusions

Although the presence of spontaneous weed-cover increased the gross primary production ( $GPP_{eco}$ ) of the olive ecosystem, weed growth reduced the net assimilation of the olive leaves ( $A_{leaf}$ ). Hence, the increase in olive ecosystem carbon uptake was not only linked to the increase in the carbon sequestration of the weed cover but also to a decrease in the carbon sequestration of the olive trees. Also, after mowing,  $A_{leaf}$  was similar for both treatments (WC and WF ecosystems) supporting the temporary nature of weed influence on olive trees fluxes. Weed growth did not affect olive leaf transpiration ( $E_{leaf}$ ). Nevertheless, a layer of mowed weeds decreased  $E_{leaf}$ , soil temperature and vapor pressure deficit in the leaves of WC ecosystem compared to WF. Therefore, weeds changed the environmental conditions towards those that decrease water losses, reducing seasonal fluctuations in water use efficiency.

Our results show that carbon sequestration at ecosystem level increases rapidly when weeds appear, whereas decreases in crop assimilation do not appear until several months afterwards. Therefore, controlling weed growth could improve the balance between erosion control, carbon sequestration and crop yield. Finally, taking into account that, (1) at the time of mowing WC had lost its ability to sequester more carbon and (2) similar values of  $A_{leaf}$  were measured only several weeks after mowing, we suggest advancing the mowing to achieve the following advantages: (i) To allow recuperation of  $A_{leaf-WC}$  at the time of fruiting to reduce differences in yield (ii) To reduce the transpiration of the weed-covered olive ecosystem in the warmer months and (iii) To increase the summer  $WUE_{eco}$ , essential in a context of climate change.

## Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

## Data availability

Data will be made available on request.

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## References

Abouziena, H.F., El-Saeid, H.M., Amin, A.B.A.E.S., 2015. Water loss by weeds: a review. *Int. J. ChemTech Res.* 7 (1), 323–336.

Aguilera, E., Lassaletta, L., Gattinger, A., Gimeno, B.S., 2013. Managing soil carbon for climate change mitigation and adaptation in Mediterranean cropping systems: a meta-analysis. *Agric. Ecosyst. Environ.* <https://doi.org/10.1016/j.agee.2013.02.003>.

Aguilera, E., Guzmán, G., Alonso, A., 2015. Greenhouse gas emissions from conventional and organic cropping systems in Spain. II. Fruit tree orchards. *Agron. Sustain. Dev.* <https://doi.org/10.1007/s13593-014-0265-y>.

Aguirre-García, S.D., Aranda-Barranco, S., Nieto, H., Serrano-Ortiz, P., Sánchez-Cañete, E.P., Guerrero-Rascado, J.L., 2021. Modelling actual evapotranspiration using a two source energy balance model with Sentinel imagery in herbaceous-free

and herbaceous-cover Mediterranean olive orchards. *Agric. For. Meteorol.* 311 <https://doi.org/10.1016/j.agrformet.2021.108692>.

Alcantara, C., Pujadas, A., Saavedra, M., 2011. Management of cruciferous cover crops by mowing for soil and water conservation in southern Spain. *Agric. Water Manag.* <https://doi.org/10.1016/j.agwat.2011.01.016>.

Allen, R. G., Pereira, L. S., Raes, D., & Smith, M. (1998). Crop evapotranspiration: Guidelines for computing crop requirements. *Irrigation and Drainage Paper No. 56*, FAO. 10.1016/j.eja.2010.12.001.

Almagro, M., de Vente, J., Boix-Fayos, C., García-Franco, N., Melgares de Aguilar, J., González, D., Solé-Benet, A., Martínez-Mena, M., 2016. Sustainable land management practices as providers of several ecosystem services under rainfed Mediterranean agroecosystems. *Mitig. Adapt. Strateg. Glob. Chang.* <https://doi.org/10.1007/s11027-013-9535-2>.

Angelopoulos, K., Dichio, B., Xiloyannis, C., 1996. Inhibition of photosynthesis in olive trees (*Olea europaea* L.) during water stress and rewatering. *J. Exp. Bot.* <https://doi.org/10.1093/jxb/47.8.1093>.

Ayyoub, A., Er-Raki, S., Khabba, S., Merlin, O., Ezzahar, J., Rodriguez, J.C., Bahlaoui, A., Chehbouni, A., 2017. A simple and alternative approach based on reference evapotranspiration and leaf area index for estimating tree transpiration in semi-arid regions. *Agric. Water Manag.* <https://doi.org/10.1016/j.agwat.2017.04.005>.

Baldocchi, D.D., 2003. Assessing the eddy covariance technique for evaluating carbon dioxide exchange rates of ecosystems: past, present and future. *Glob.Chang. Biol.* <https://doi.org/10.1046/j.1365-2486.2003.00629.x>.

Baldocchi, D.D., 2020. How eddy covariance flux measurements have contributed to our understanding of Global Change Biology. *Glob. Chang. Biol.* 26 (1) <https://doi.org/10.1111/gcb.14807>.

Baraibar, B., Carrión, E., Recasens, J., Westerman, P.R., 2011. Unravelling the process of weed seed predation: developing options for better weed control. *Biol. Control.* <https://doi.org/10.1016/j.biocontrol.2010.09.010>.

Basche, A., DeLonge, M., 2017. The Impact of continuous living cover on soil hydrologic properties: a meta-analysis. *Soil Sci. Soc. Am. J.* <https://doi.org/10.2136/sssaj2017.03.0077>.

Basche, A.D., DeLonge, M.S., 2019. Comparing infiltration rates in soils managed with conventional and alternative farming methods: a meta-analysis. *PLoS One.* <https://doi.org/10.1371/journal.pone.0215702>.

Bedbabis, S., Rouina, B.B., Camposeo, S., Clodoveo, M.L., Gallotta, A., Palasciano, M., Ferrara, G., 2017. Alternate bearing affects nutritional status and net assimilation rate of an irrigated olive grove under arid conditions. *Acta Sci. Pol. Hortorum Cultus* 16 (2). <https://doi.org/10.24326/asphc.2017.2.0>.

Ben Ahmed, C., Ben Rouina, B., Boukhris, M., 2007. Effects of water deficit on olive trees cv. Chemlali under field conditions in arid region in Tunisia. *Sci. Hortic.* <https://doi.org/10.1016/j.scienta.2007.03.020>.

Bertolla, C., Caruso, G., Gucci, R., 2014. Seasonal changes in soil respiration rates in olive orchards. *Acta Hortic.* <https://doi.org/10.17660/ActaHortic.2014.1057.30>.

Blanco-Canqui, H., Shaver, T.M., Lindquist, J.L., Shapiro, C.A., Elmore, R.W., Francis, C. A., Hergert, G.W., 2015. Cover crops and ecosystem services: insights from studies in temperate soils. *Agron. J.* <https://doi.org/10.2134/agronj15.0086>.

Boese, S., Jung, M., Carvalhais, N., Teuling, A.J., Reichstein, M., 2019. Carbon–water flux coupling under progressive drought. *Biogeosciences* 16 (13), 2557–2572. <https://doi.org/10.5194/bg-16-2557-2019>.

Brilli, L., Gioli, B., Toscano, P., Moriondo, M., Zaldei, A., Cantini, C., Ferrise, R., Bindi, M., 2016. Rainfall regimes control C-exchange of Mediterranean olive orchard. *Agric. Ecosyst. Environ.* <https://doi.org/10.1016/j.agee.2016.09.006>.

Carrer, D., Pique, G., Ferlicq, M., Ceamanos, X., Ceschia, E., 2018. What is the potential of cropland albedo management in the fight against global warming? A case study based on the use of cover crops. *Environ. Res. Lett.* 13 (4) <https://doi.org/10.1088/1748-9326/aab650>.

Castro, J., Fernández-Ondoño, E., Rodríguez, C., Lallena, A.M., Sierra, M., Aguilar, J., 2008. Effects of different olive-grove management systems on the organic carbon and nitrogen content of the soil in Jaén (Spain). *Soil Tillage Res.* <https://doi.org/10.1016/j.still.2007.10.002>.

Chamizo, S., Serrano-Ortiz, P., López-Ballesteros, A., Sánchez-Cañete, E.P., Vicente-Vicente, J.L., Kowalski, A.S., 2017. Net ecosystem CO<sub>2</sub> exchange in an irrigated olive orchard of SE Spain: influence of weed cover. *Agric. Ecosyst. Environ.* 239, 51–64. <https://doi.org/10.1016/j.agee.2017.01.016>.

Chebbi, W., Boulet, G., Le Dantec, V., Lili Chabaane, Z., Fanise, P., Mougnot, B., Ayari, H., 2018. Analysis of evapotranspiration components of a rainfed olive orchard during three contrasting years in a semi-arid climate. *Agric. For. Meteorol.* <https://doi.org/10.1016/j.agrformet.2018.02.020>.

Connor, D.J., 2005. Adaptation of olive (*Olea europaea* L.) to water-limited environments. *Aust. J. Agric. Res.* <https://doi.org/10.1071/AR05169>.

de Brogniez, D., Ballabio, C., Stevens, A., Jones, R.J.A., Montanarella, L., van Wesemael, B., 2015. A map of the topsoil organic carbon content of Europe generated by a generalized additive model. *Eur. J. Soil Sci.* 66 (1) <https://doi.org/10.1111/ejss.12193>.

Droogers, P., 2000. Estimating actual evapotranspiration using a detailed agro-hydrological model. *J. Hydrol.* [https://doi.org/10.1016/S0022-1694\(99\)00198-5](https://doi.org/10.1016/S0022-1694(99)00198-5).

FAOSTAT (2019). Food and agriculture organization of the united nations statistical dataset.

Fernández, J.E., Moreno, F., 1999. Water use by the olive tree. *J. Crop Prod.* [https://doi.org/10.1300/J144v02n02\\_05](https://doi.org/10.1300/J144v02n02_05).

Ferreira, I.Q., Arrobas, M., Claro, A.M., Rodrigues, M.A., 2013. Soil management in rainfed olive orchards may result in conflicting effects on olive production and soil fertility. *Span. J. Agric. Res.* <https://doi.org/10.5424/sjar/2013112-3501>.

Fratini, G., Ibrom, A., Arriga, N., Burba, G., Papale, D., 2012. Relative humidity effects on water vapour fluxes measured with closed-path eddy-covariance systems with short

- sampling lines. *Agric. For. Meteorol.* <https://doi.org/10.1016/j.agrformet.2012.05.018>.
- García-Ruiz, J.M., Nadal-Romero, E., Lana-Renault, N., Beguería, S., 2013. Erosion in Mediterranean landscapes: changes and future challenges. *Geomorphology*. <https://doi.org/10.1016/j.geomorph.2013.05.023>.
- Gómez, J.A., Romero, P., Giráldez, J.V., Fereres, E., 2004. Experimental assessment of runoff and soil erosion in an olive grove on a Vertic soil in southern Spain as affected by soil management. *Soil Use Manag.* <https://doi.org/10.1079/sum2004275>.
- Gómez, J.A., Guzmán, M.G., Giráldez, J.V., Fereres, E., 2009. The influence of cover crops and tillage on water and sediment yield, and on nutrient, and organic matter losses in an olive orchard on a sandy loam soil. *Soil Tillage Res.* <https://doi.org/10.1016/j.still.2009.04.008>.
- González-Sánchez, E.J., Ordóñez-Fernández, R., Carbonell-Bojollo, R., Veroz-González, O., Gil-Ribes, J.A., 2012. Meta-analysis on atmospheric carbon capture in Spain through the use of conservation agriculture. *Soil Tillage Res.* <https://doi.org/10.1016/j.still.2012.03.001>.
- Gregoriou, K., Pontikis, K., Vemmos, S., 2007. Effects of reduced irradiance on leaf morphology, photosynthetic capacity, and fruit yield in olive (*Olea europaea* L.). *Photosynthetica* 45 (2). <https://doi.org/10.1007/s11099-007-0029-x>.
- Hagidimitriou, M., Pontikis, C.A., 2005. Seasonal changes in CO<sub>2</sub> assimilation in leaves of five major Greek olive cultivars. *Sci. Hortic.* <https://doi.org/10.1016/j.scienta.2004.07.011>.
- Haruna, S.I., Nkongolo, N.V., Anderson, S.H., Eivazi, F., Zaibon, S., 2018. *In situ* infiltration as influenced by cover crop and tillage management. *J. Soil Water Conserv.* <https://doi.org/10.2489/jswc.73.2.164>.
- Higgins, S.S., Larsen, F.E., Bendel, R.B., Radamaker, G.K., Bassman, J.H., Bidlake, W.R., Al Wir, A., 1992. Comparative gas exchange characteristics of potted, glasshouse-grown almond, apple, fig, grape, olive, peach and Asian pear. *Sci. Hortic.* [https://doi.org/10.1016/0304-4238\(92\)90032-8](https://doi.org/10.1016/0304-4238(92)90032-8).
- IFAPA. (26 de Junio de 2020). Instituto Andaluz de Investigación y Formación Agraria, Pesquera, Alimentaria y de la Producción Ecológica (IFAPA). Estaciones Agroclimáticas: Obtenido de: <https://www.juntadeandalucia.es/agriculturaypesca/ifapa/ria/servlet/FrontController>.
- Kim, N., Zabaloy, M.C., Guan, K., Villamil, M.B., 2020. Do cover crops benefit soil microbiome? A meta-analysis of current research. *Soil Biol. Biochem.* <https://doi.org/10.1016/j.soilbio.2019.107701>.
- Krom, M.D., 1980. Spectrophotometric determination of ammonia: a study of a modified berthelot reaction using salicylate and dichloroisocyanurate. *Analyst* 105 (1249). <https://doi.org/10.1039/an9800500305>.
- Lal, R., 2019. Accelerated soil erosion as a source of atmospheric CO<sub>2</sub>. *Soil Tillage Res.* <https://doi.org/10.1016/j.still.2018.02.001>.
- Larbi, A., Vázquez, S., El-Jendoubi, H., Msallem, M., Abadía, J., Abadía, A., Morales, F., 2015. Canopy light heterogeneity drives leaf anatomical, eco-physiological, and photosynthetic changes in olive trees grown in a high-density plantation. *Photosynth. Res.* <https://doi.org/10.1007/s11220-014-0052-2>.
- Lee, H., Lautenbach, S., Nieto, A.P.G., Bondeau, A., Cramer, W., Geijendorffer, I.R., 2019. The impact of conservation farming practices on Mediterranean agro-ecosystem services provisioning—a meta-analysis. *Reg. Environ. Chang.* <https://doi.org/10.1007/s10113-018-1447-y>.
- López-Bernal, Á., García-Tejera, O., Vega, V.A., Hidalgo, J.C., Testi, L., Orgaz, F., Villalobos, F.J., 2015. Using sap flow measurements to estimate net assimilation in olive trees under different irrigation regimes. *Irrig. Sci.* 33 (5) <https://doi.org/10.1007/s00271-015-0471-7>.
- Mariotti, A., Pan, Y., Zeng, N., Alessandri, A., 2015. Long-term climate change in the Mediterranean region in the midst of decadal variability. *Clim. Dyn.* <https://doi.org/10.1007/s00382-015-2487-3>.
- Marland, G., Garten, C.T., Post, W.M., West, T.O., 2004. Studies on enhancing carbon sequestration in soils. *Energy*. <https://doi.org/10.1016/j.energy.2004.03.066>.
- Marquez-García, F., Gonzalez-Sanchez, E.J., Castro-García, S., Ordoñez-Fernandez, R., 2013. Improvement of soil carbon sink by cover crops in olive orchards under semiarid conditions. Influence of the type of soil and weed. *Span. J. Agric. Res.* <https://doi.org/10.5424/sjar/2013112-3558>.
- Martinez-Mena, M., Lopez, J., Almagro, M., Boix-Fayos, C., Albaladejo, J., 2008. Effect of water erosion and cultivation on the soil carbon stock in a semiarid area of South-East Spain. *Soil Tillage Res.* <https://doi.org/10.1016/j.still.2008.01.009>.
- Martinez-Mena, M., Carrillo-López, E., Boix-Fayos, C., Almagro, M., García Franco, N., Díaz-Pereira, E., Montoya, I., de Vente, J., 2020. Long-term effectiveness of sustainable land management practices to control runoff, soil erosion, and nutrient loss and the role of rainfall intensity in Mediterranean rainfed agroecosystems. *Catena*. <https://doi.org/10.1016/j.catena.2019.104352>.
- Mauder, M., Cuntz, M., Drüe, C., Graf, A., Rebmann, C., Schmid, H.P., Schmidt, M., Steinbrecher, R., 2013. A strategy for quality and uncertainty assessment of long-term eddy-covariance measurements. *Agric. For. Meteorol.* 169 <https://doi.org/10.1016/j.agrformet.2012.09.006>.
- Mingorance, M., Barahona, E., Fernández-Gálvez, J., 2007. Guidelines for improving organic carbon recovery by the wet oxidation method. *Chemosphere* 68 (3), 409–413. <https://doi.org/10.1016/j.chemosphere.2007.01.021>.
- Moncrieff, J., Clement, R., Finnigan, J., Meyers, T., 2006. Averaging, detrending, and filtering of eddy covariance time series. *Handbook of Micrometeorology*. [https://doi.org/10.1007/1-4020-2265-4\\_2](https://doi.org/10.1007/1-4020-2265-4_2).
- Montenegro, A.A.A., Abrantes, J.R.C.B., De Lima, J.L.M.P., Singh, V.P., Santos, T.E.M., 2013. Impact of mulching on soil and water dynamics under intermittent simulated rainfall. *Catena*. <https://doi.org/10.1016/j.catena.2013.03.018>.
- Moriana, A., Villalobos, F.J., Fereres, E., 2002. Stomatal and photosynthetic responses of olive (*Olea europaea* L.) leaves to water deficits. *Plant Cell Environ.* 25 (3) <https://doi.org/10.1046/j.0016-8025.2001.00822.x>.
- Moriana, A., Orgaz, F., Pastor, M., Fereres, E., 2003. Yield responses of a mature olive orchard to water deficits. *J. Am. Soc. Hortic. Sci.* <https://doi.org/10.21273/jahs.128.3.0425>.
- Morugán-Coronado, A., Linares, C., Gómez-López, M.D., Faz, Á., Zornoza, R., 2020. The impact of intercropping, tillage and fertilizer type on soil and crop yield in fruit orchards under Mediterranean conditions: a meta-analysis of field studies. *Agric. Syst.* 178 (December), 102736 <https://doi.org/10.1016/j.agry.2019.102736>.
- Nardino, M., Pernice, F., Rossi, F., Georgiadis, T., Facini, O., Motisi, A., Drago, A., 2013. Annual and monthly carbon balance in an intensively managed Mediterranean olive orchard. *Photosynthetica*. <https://doi.org/10.1007/s11099-012-0079-6>.
- Nicholls, C.I., Altieri, M.A., 2013. Plant biodiversity enhances bees and other insect pollinators in agroecosystems. A review. *Agron. Sustain. Dev.* <https://doi.org/10.1007/s13593-012-0092-y>.
- Nieto, O.M., Castro, J., Fernández-Ondoño, E., 2013. Conventional tillage versus cover crops in relation to carbon fixation in Mediterranean olive cultivation. *Plant Soil*. <https://doi.org/10.1007/s11104-012-1395-0>.
- Novara, A., Cerda, A., Barone, E., Gristina, L., 2021. Cover crop management and water conservation in vineyard and olive orchards. *Soil Tillage Res.* 208 <https://doi.org/10.1016/j.still.2020.104896>.
- Orgaz, F., Testi, L., Villalobos, F.J., Fereres, E., 2006. Water requirements of olive orchards-II: determination of crop coefficients for irrigation scheduling. *Irrig. Sci.* <https://doi.org/10.1007/s00271-005-0012-x>.
- Orgaz, F., Villalobos, F.J., Testi, L., Fereres, E., 2007. A model of daily mean canopy conductance for calculating transpiration of olive canopies. *Funct. Plant Biol.* <https://doi.org/10.1071/FP06306>.
- Paço, T.A., Pôças, I., Cunha, M., Silvestre, J.C., Santos, F.L., Paredes, P., Pereira, L.S., 2014. Evapotranspiration and crop coefficients for a super intensive olive orchard. An application of SIMDualKc and METRIC models using ground and satellite observations. *J. Hydrol.* <https://doi.org/10.1016/j.jhydrol.2014.09.075>.
- Palese, A.M., Pergola, M., Favia, M., Xiloyannis, C., Celano, G., 2013. A sustainable model for the management of olive orchards located in semi-arid marginal areas: some remarks and indications for policy makers. *Environ. Sci. Policy*. <https://doi.org/10.1016/j.envsci.2012.11.001>.
- Palomo, M.J., Moreno, F., Fernández, J.E., Díaz-Espejo, A., Girón, I.F., 2002. Determining water consumption in olive orchards using the water balance approach. *Agric. Water Manag.* [https://doi.org/10.1016/S0378-3774\(01\)00182-2](https://doi.org/10.1016/S0378-3774(01)00182-2).
- Pastor, M., Castro, J., Humanes, M.D., Muñoz, J., 2001. Sistemas de manejo del suelo en olivar de Andalucía. *Edafología* 8, 75–98.
- Pérez-Priego, O., Testi, L., Kowalski, A.S., Villalobos, F.J., Orgaz, F., 2014. Aboveground respiratory CO<sub>2</sub> effluxes from olive trees (*Olea europaea* L.). *Agrofor. Syst.* <https://doi.org/10.1007/s10457-014-9672-y>.
- Poeplau, C., Don, A., 2015. Carbon sequestration in agricultural soils via cultivation of cover crops - a meta-analysis. *Agric. Ecosyst. Environ.* <https://doi.org/10.1016/j.agee.2014.10.024>.
- Pou, A., Gullás, J., Moreno, M., Tomás, M., Medrano, H., Cifre, J., 2011. Cover cropping in *Vitis vinifera* L. cv. Manto Negro vineyards under Mediterranean conditions: effects on plant vigour, yield and grape quality. *OENO One* 45 (4), 223–234. <https://doi.org/10.20870/oeno-one.2011.45.4.1501>.
- Proietti, P., Famiani, F., 2002. Diurnal and seasonal changes in photosynthetic characteristics in different olive (*Olea europaea* L.) cultivars. *Photosynthetica*. <https://doi.org/10.1023/A:1021329220613>.
- Proietti, P., Nasini, L., Ilarioni, L., 2012. Photosynthetic behavior of Spanish Arbequina and Italian Maurino olive (*Olea europaea* L.) cultivars under super-intensive grove conditions. *Photosynthetica* 50 (2). <https://doi.org/10.1007/s11099-012-0025-7>.
- Ramos, M.E., Robles, A.B., Sánchez-Navarro, A., González-Rebollar, J.L., 2011. Soil responses to different management practices in rainfed orchards in semiarid environments. *Soil Tillage Res.* <https://doi.org/10.1016/j.still.2010.11.007>.
- Rapoport, H.F., Pérez-López, D., Hammami, S.B.M., Agüera, J., Moriana, A., 2013. Fruit pit hardening: physical measurement during olive fruit growth. *Ann. Appl. Biol.* 163 (2) <https://doi.org/10.1111/aab.12046>.
- Reichstein, M., Falge, E., Baldocchi, D., Papale, D., Aubinet, M., Berbigier, P., Bernhofer, C., Buchmann, N., Gilmanov, T., Granier, A., Grünwald, T., Havránková, K., Ilvesniemi, H., Janous, D., Knohl, A., Laurila, T., Lohila, A., Loustau, D., Matteucci, G., Valentini, R., 2005. On the separation of net ecosystem exchange into assimilation and ecosystem respiration: review and improved algorithm. *Glob. Chang. Biol.* <https://doi.org/10.1111/j.1365-2486.2005.001002.x>.
- Repullo-Ruibérriz de Torres, M.A., Ordóñez-Fernández, R., Giráldez, J.V., Márquez-García, J., Laguna, A., Carbonell-Bojollo, R., 2018. Efficiency of four different seeded plants and native vegetation as cover crops in the control of soil and carbon losses by water erosion in olive orchards. *Land Degrad. Dev.* <https://doi.org/10.1002/ldr.3023>.
- Rodríguez Martín, J.A., Álvaro-Fuentes, J., Gonzalo, J., Gil, C., Ramos-Miras, J.J., Grau Corbí, J.M., Boluda, R., 2016. Assessment of the soil organic carbon stock in Spain. *Geoderma*. <https://doi.org/10.1016/j.geoderma.2015.10.010>.
- Rothfuss, Y., Biron, P., Braud, I., Canale, L., Durand, J.L., Gaudet, J.P., Richard, P., Vauclin, M., Bariac, T., 2010. Partitioning evapotranspiration fluxes into soil evaporation and plant transpiration using water stable isotopes under controlled conditions. *Hydrol. Process.* 24 (22) <https://doi.org/10.1002/hyp.7743>.
- Sastre, B., Marques, M.J., García-Díaz, A., Bienes, R., 2018. Three years of management with cover crops protecting sloping olive groves soils, carbon and water effects on gypsiferous soil. *Catena*. <https://doi.org/10.1016/j.catena.2018.07.003>.
- Schmidt, R., Mitchell, J., Scow, K., 2019. Cover cropping and no-till increase diversity and symbiotroph:saprotroph ratios of soil fungal communities. *Soil Biol. Biochem.* <https://doi.org/10.1016/j.soilbio.2018.11.010>.
- Sierra, M., Martínez, F.J., Braojos, V., Romero-Freire, A., Ortiz-Bernad, I., Martín, F.J., 2016. Chemical stabilization of organic carbon in agricultural soils in a semi-arid

- region (SE Spain). *J. Agric. Sci.* 154 (1), 87–97. <https://doi.org/10.1017/S002185961500012X>.
- Slaughter, D.C., Giles, D.K., Downey, D., 2008. Autonomous robotic weed control systems: a review. *Comput. Electron. Agric.* <https://doi.org/10.1016/j.compag.2007.05.008>.
- Sofo, A., Nuzzo, V., Palese, A.M., Xiloyannis, C., Celano, G., Zukowskyj, P., Dichio, B., 2005. Net CO<sub>2</sub> storage in mediterranean olive and peach orchards. *Sci. Hortic.* <https://doi.org/10.1016/j.scienta.2005.06.001>.
- Sofo, A., Dichio, B., Montanaro, G., Xiloyannis, C., 2009. Shade effect on photosynthesis and photoinhibition in olive during drought and rewatering. *Agric. Water Manag.* 96 (8) <https://doi.org/10.1016/j.agwat.2009.03.004>.
- Sokos, C.K., Mamolos, A.P., Kalburtji, K.L., Birtsas, P.K., 2013. Farming and wildlife in Mediterranean agroecosystems. *J. Nat. Conserv.* <https://doi.org/10.1016/j.jnc.2012.11.001>.
- Tanasijevic, L., Todorovic, M., Pereira, L.S., Pizzigalli, C., Lionello, P., 2014. Impacts of climate change on olive crop evapotranspiration and irrigation requirements in the Mediterranean region. *Agric. Water Manag.* <https://doi.org/10.1016/j.agwat.2014.05.019>.
- Testi, L., Villalobos, F.J., Orgaz, F., 2004. Evapotranspiration of a young irrigated olive orchard in southern Spain. *Agric. For. Meteorol.* <https://doi.org/10.1016/j.agrformet.2003.08.005>.
- Testi, L., Orgaz, F., Villalobos, F.J., 2006. Variations in bulk canopy conductance of an irrigated olive (*Olea europaea* L.) orchard. *Environ. Exp. Bot.* 55 (1–2) <https://doi.org/10.1016/j.envexpbot.2004.09.008>.
- Testi, L., Orgaz, F., Villalobos, F., 2008. Carbon exchange and water use efficiency of a growing, irrigated olive orchard. *Environ. Exp. Bot.* <https://doi.org/10.1016/j.envexpbot.2007.11.006>.
- Torres-Ruiz, J.M., Fernández, J.E., Girón, I.F., Romero, R., Jiménez-Bocanegra, J.A., García Tejero, I., Martín-Palomo, M.J., 2012. Determining evapotranspiration in an olive orchard in Southwest Spain. *Acta Hort.* 949 <https://doi.org/10.17660/ActaHortic.2012.949.36>.
- Turrini, A., Caruso, G., Avio, L., Gennai, C., Palla, M., Agnolucci, M., Tomei, P.E., Giovannetti, M., Gucci, R., 2017. Protective green cover enhances soil respiration and native mycorrhizal potential compared with soil tillage in a high-density olive orchard in a long term study. *Appl. Soil Ecol.* <https://doi.org/10.1016/j.apsoil.2017.04.001>.
- Vicente-Vicente, J.L., García-Ruiz, R., Francaviglia, R., Aguilera, E., Smith, P., 2016. Soil carbon sequestration rates under Mediterranean woody crops using recommended management practices: a meta-analysis. *Agric. Ecosyst. Environ.* <https://doi.org/10.1016/j.agee.2016.10.024>.
- Vickers, D., Mahrt, L., 1997. Quality control and flux sampling problems for tower and aircraft data. *J. Atmos. Ocean. Technol.* [https://doi.org/10.1175/1520-0426\(1997\)014<0512:QCAFSP>2.0.CO;2](https://doi.org/10.1175/1520-0426(1997)014<0512:QCAFSP>2.0.CO;2).
- Villalobos, F.J., Testi, L., Hidalgo, J., Pastor, M., Orgaz, F., 2006. Modelling potential growth and yield of olive (*Olea europaea* L.) canopies. *Eur. J. Agron.* <https://doi.org/10.1016/j.eja.2005.10.008>.
- Villalobos, F.J., Perez-Priego, O., Testi, L., Morales, A., Orgaz, F., 2012. Effects of water supply on carbon and water exchange of olive trees. *Eur. J. Agron.* <https://doi.org/10.1016/j.eja.2012.02.004>.
- Wellburn, A.R., 1994. The spectral determination of chlorophylls a and b, as well as total carotenoids, using various solvents with spectrophotometers of different resolution. *J. Plant Physiol.* [https://doi.org/10.1016/S0176-1617\(11\)81192-2](https://doi.org/10.1016/S0176-1617(11)81192-2).
- Yunusa, I.A.M., Zeppel, M.J.B., Nuberg, I.K., 2008. Water-use efficiency reflects management practices in Australian olive groves. *J. Hortic. Sci. Biotechnol.* 83 (2) <https://doi.org/10.1080/14620316.2008.11512374>.